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A preliminary report of amphibian mortality patterns on railways

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Abstract. In contrast to road mortality, little is known about amphibian railroad mortality. The aim of this study was to quantify amphibian mortality along a railway line as well as to investigate the relationship between the availability of breeding sites in the surrounding habitats and the monthly variation of amphibian railway mortality. The study was conducted from April to July 2011 along 45 km of the railway line Kraków - Tarnów (Poland, Małopolska province). Three species were affected by railway mortality: *Bufo bufo, Rana temporaria* and *Pelophylax* kl. *esculentus*. Most dead individuals (77%) were adult common toads. The largest number (14) of amphibian breeding sites was located in the most heterogeneous habitats (woodland and rural areas), which coincides with the sectors of highest amphibian mortality (42% of all accidents). As in the case of roads, spring migration is the period of highest amphibian mortality (87% of all accidents) on railroads. Our findings suggest that railroad mortality depends on the agility of the species, associated primarily with the ability to overcome the rails.

Keywords. Habitat effect, seasonality effect, common toad, Poland.

One of the main consequences of urbanization is the construction of new communication networks, e.g. linear infrastructures such as roads and railways. Roads are physical barriers to animal migration, which may have negative consequences both in terms of animal mortality and habitat fragmentation (Andrews and Gibbons, 2005) and, in turn, may lead to isolation of populations through reduced movement and gene flow (Gibbs, 1998; St. Clair, 2003). Among vertebrates, amphibians are the most affected by these threats (Stuart et al., 2004). Their requirement of aquatic habitats and reproduction-dependent seasonal migrations make them particularly vulnerable to the negative impact of road traffic (Hels and Buchwald, 2001; Hamer and McDonnell, 2008). Apart from roads, railways may also act as migratory barriers and thus negatively affect amphibian populations (Berthoud and Antoniazza, 1998; Ray et al., 2002). To date, the impact of railways on amphibians has not been established and, in contrast to the issue of amphibian road mortality (Carr and Fahrig, 2001; Mazerolle, 2004; Sirello, 2008; Sutherland et al., 2010), data on amphibian mortality due to the presence of railways are very scarce (Berthoud and Antoniazza, 1998; Vos et al., 2001; Reshetylo and Briggs, 2010). The aim of this study was to quantify amphibian mortality along a railway line and to investigate the effect of the surrounding habitat and the seasonal variation of railway mortality of amphibians. The study was conducted along 45 km of the line Podłęże - Biadoliny (direction Kraków - Tarnów, southern Poland) (Fig. 1). The railroad is constituted by two rail lines that split into several others where large stations occur. The track spacing is 1.435 m wide, and the height of the rail profile is 0.172 m. The substrate of the tracks is made of stones. The average daily number of trains running on this route in both directions is about 60. The trains run between 3:00 am and 23:00 pm. The average frequency of trains is 2-3 trains / h, increasing up to 3-4 trains / h from 14:00 to 20:00 (due to a lack of data, freight trains were not included). The study site included highly urbanized and agricultural

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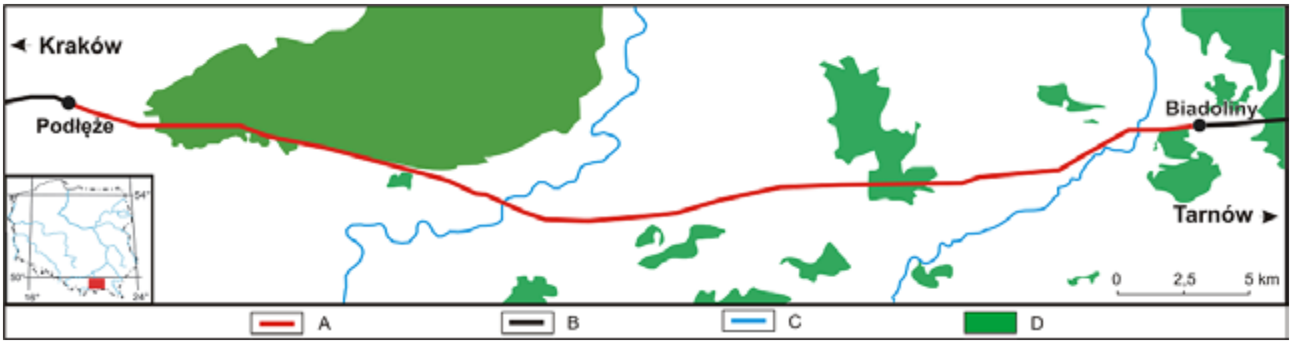

Fig. 1. Location of the surveyed transect in Poland. Legend: A - surveyed transect of railway line, B - further railway line, C - rivers, D - forests.

areas, grasslands and forests. Numerous ditches, oxbows and wetlands, as well as some larger water bodies, such as fish ponds, occur near the railway line and constitute potential breeding habitat for amphibians. The stretch was divided into 30 transects associated with different types of habitat. Five types of transects were established: 'woodland' transects with woodland on both sides of the railroad (six transects; total length: 11.35 km), 'woodland and rural areas' transects with woodland on one side of the railroad and rural areas on the other side (four transects; total length: 6.16 km), 'rural areas' transects with rural areas on both sides of the railroad (10 transects; total length: 14.74 km), 'open areas' transects with open, natural areas (eight transects; total length: 9.8 km) and 'urban areas' transects (two transects; total length: 3.25 km). The study was conducted from April to July 2011. In April and May each transect was monitored twice a month, while in June and July, once a month. All transects were surveyed on foot. The duration of each survey was 1 to 3 hours. The surveys were conducted from the morning until the evening (often three or four transects a day), usually in sunny and dry weather. All findings of dead amphibians were georeferenced, photographed, and information on amphibian species and age (juvenile or adult) were taken. This detailed information ensured that we avoided recounting of dead individuals, even though we did not remove dead amphibians from the rails. Additionally, the presence of dead reptiles was registered. A buffer zone of about 150 m on both sides of the railway was monitored for the presence of amphibians and potential reproductive sites at the same time as the railway mortality surveys. The inspections consisted of searching through all ditches, pools, puddles and water bodies, their edges and vicinities. The water reservoirs were also dipnetted. All individuals were released after identification in the field. The determination of amphibian presence was based on direct observations of adults and juveniles, as well as on observations of spawn, larvae and male mating calls. All observed green frogs were classified as *Pelophylax* kl. *esculentus*. Chi square tests were used to assess differences in railroad mortality depending on habitat type and month. Additionally, differences in number of breeding sites in different habitat types were assessed. The analysis included only breeding sites of species affected by railroad mortality. Then, differences between pairs of habitat types in respect to railroad mortality and breeding site abundance were tested. Spearman's correlation was used to measure the association between the number of dead specimens found on the railroad for each species with the number of reproductive sites found in the buffer zone.

Within the study area we found the following species (the number of breeding sites is given in parentheses): the agile frog *Rana dalmatina* (23 sites), the common frog *R. temporaria* (7 sites), the moor frog *R. arvalis* (1 site), the green frogs *Pelophylax* kl. *esculentus* (43 sites), the European tree frog *Hyla arborea* (1 site), the fire-bellied toad *Bombina bombina* (10 sites), the common toad *Bufo bufo* (5 sites), the great crested newt *Triturus cristatus* (4 sites), and the smooth newt *Lissotriton vulgaris* (1 site). A total of 62 dead individuals of three species (*B. bufo*, *R. temporaria* and *P.* kl *esculentus*) were found within the area of the railway tracks. Seven frog specimens were not identified. Most dead amphibians were adult common toads (77%), and a large proportion of dead frogs (73%) were juveniles. The transect differed in terms of amphibian mortality ($\chi^2 = 54.4$, df = 4, p-value < 0.001): the majority of the amphibian mortality occurred in woodland and rural areas (Fig. 2, Table 1). The buffer zone areas (habitat types) varied in terms of amphibian breeding site abundance ($\chi^2 = 10.8$, df = 4, p-value < 0.05). Most of the breeding sites (of amphibians affected by railroad mortality) were located in the 'woodland and rural areas' type (Table 2). The number of dead speci-

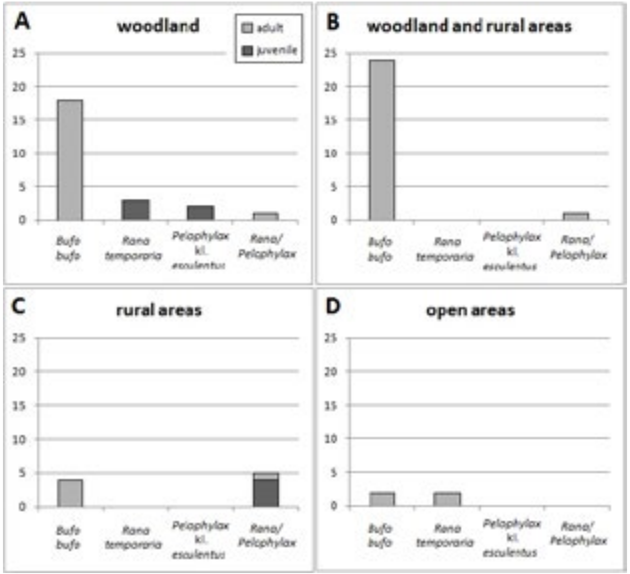

Fig. 2. Number of dead individuals (including their age) found in different types of habitat: woodland (A), woodland and rural areas (B), rural areas (C) and open areas (D).

mens and the number of reproductive sites occurring in the habitat types was not significantly correlated for any of the species. However, this association is present if all dead frogs (*Rana temporaria*, *Pelophylax* kl.*esculentus* and unspecified *Rana*/*Pelophylax*) are taken together (R = 0.436, p-value < 0.05). There is also a significant relationship between dead *B. bufo* and breeding sites abundance (R = 0.458, p-value < 0.001), if three breeding sites situated outside the buffer zone (up to 1.3 km in a straight line from the tracks) (Budzik K. M., pers. inf.) are taken into account. The majority of dead amphibians were found at the beginning of the reproductive season ($\chi^2 = 128.2$, df = 3, p-value < 0.001; Fig. 3). Most dead amphibians found in April were spatially clustered, while in the following months the specimens were scattered. Many of the toads (58%) were found within the railroad tracks and their remains were fragmented. The remaining individuals, as well as other dead amphibians, were not mechanically damaged. All dead frogs were found outside of the railroad track. Additionally, in May we found one road-killed fire-bellied toad under one of the rail viaducts. We found six dead grass snakes (*Natrix natrix*), one of which was found near a dead common toad (Fig. 4).

To our knowledge, this study - despite being largely exploratory - reports the first empirical data on amphibian railway mortality. Our results show that railway mortality is a real threat for amphibians, an issue that requires deeper evaluation for conservation planning. The amphibians found in the study area are common in this region of Poland (Głowaciński and Rafiński, 2003). Furthermore, two of the three species affected by railroad mortality (*B. bufo, R. temporaria*), are among the most common European amphibians, for which there is evidence of great road-mortality (Orłowski, 2007; Bonardi et al., 2011; Matos et al., 2012). The high number of amphibians killed along woodland and rural areas is likely associated with the abundance of breeding sites in these types of habitats. However, the results predominantly relate to the common toad, therefore they are highly conditioned by this species, which typically inhabits heterogeneous habitats (Pavignano et al., 1990).

Table 1. Chi-square test comparing the railroad mortality between each pair of habitat types. "-" refers to low expected frequencies, test is not applicable.

	Woodland	Woodland and Rural	Rural	Open
Woodland and Rural	$\chi^2 = 5.4$, df=1, p < 0.05			
Rural	$\chi^2 = 11.5$, df=1, p < 0.001	$\chi^2 = 31.7$, df=1, p < 0.001		
Open	$\chi^2 = 11.5$, df=1, p < 0.001	$\chi^2 = 27.7$, df=1, p < 0.001	-	
Urban	$\chi^2 = 6.9$, df=1, p < 0.01	$\chi^2 = 13.2$, df=1, p < 0.001	-	-

Table 2. Chi-square test comparing the number of *B. bufo*, *R. temporaria*, and *P.* kl *esculentus* breeding sites between each pair of habitat types. "-" refers to low expected frequencies, test is not applicable. NS: non significant p-value.

	Woodland	Woodland and Rural	Rural	Open
Woodland and Rural	$\chi^2 = 1.7$, df=1, NS			
Rural	$\chi^2 = 2.7$, df=1, NS	$\chi^2 = 8.4$, df=1, p < 0.01		
Open	$\chi^2 = 1.1$, df=1, NS	$\chi^2 = 4.8$, df=1, p < 0.05	-	
Urban	$\chi^2 = 1.3$, df=1, NS	$\chi^2 = 3.4$, df=1, NS	-	-

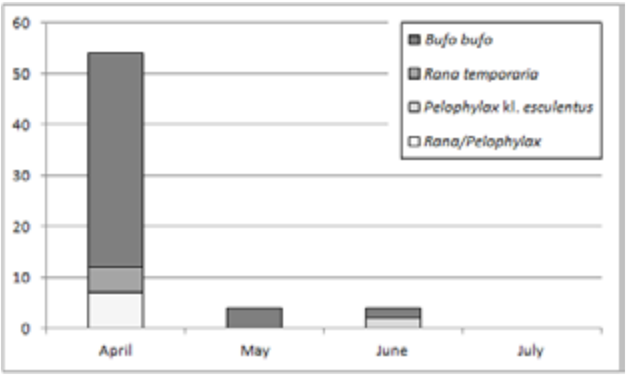

Fig. 3. Number of dead individuals found in each month of the survey.

Fig. 4. Dead common toad (*Bufo bufo*, arrow) on the tracks with dead grass snake (*Natrix natrix*). Photo by K.M. Budzik.

Most dead amphibians were spatially clustered in April but this result appears conditioned by the large number of common toads and their mass migrations to breeding sites. Toads were scattered after the breeding season, suggesting seasonal migrations towards feeding grounds. Additionally, as in the case of roads (Hels and Buchwald, 2001; Hamer and McDonnell, 2008), spring migration seems to be the period of highest amphibian mortality on railroad tracks. Undetermined frogs were probably representatives of the common frog or moor frog, which awakened from hibernation in April. The peak of green frog mortality in June may indicate dispersal in search of new habitats because of the gradual drying of habitat in ditches alongside the railroad tracks. The fragmentation of the remains of common toads clearly suggests that the direct cause of death was collision with a train. The short limbs of these animals reduce their ability of overcoming barriers such as rails. In addition, numerous studies have shown that amphibians are likely to remain immobile if faced with an approaching light (Cornell and Hailman, 1984; Mazerolle et al., 2005). Thus, it is possible that common toad activity can be disturbed under train light, increasing the risk of mortality. The toads that were not damaged, but trapped inside the track, probably died of dehydration. Because dead frog individuals were not mechanically damaged, we suppose that they were probably hit by a train while trying to overcome the rails. The majority of dead frogs were juveniles. We suggest that most adults, able to hop farther and faster than the juveniles, may migrate more successfully. We did not find any dead individuals of the agile frog, the European tree frog, the fire-bellied toad or newts. As regards the latter, small-sized species may avoid the tracks because they are unable to cross them. To successfully migrate, their only option may be to avoid the rails and rather move along the viaducts: this suggestion is worthy of further investigation. However, this result may also be due to a sampling issue: on the one hand, small-sized amphibians dry up faster; on the other hand they may be crushed by a train; either way, this would make them very difficult to detect (Dodd et al., 2004; Mazerolle et al., 2005). There is also the possibility that small-sized amphibians may migrate through a gap under the railway. The agile frog and probably the other frogs seem to successfully cross the rails, probably thanks to their jumping ability. Railroad mortality seems to depend on physical features (such as body size, limb length) and may be associated with the agility of the species. In the case of roads, agility was related mainly to velocity of the individual (Schlupp and Podloucky, 1994; Hels and Buchwald, 2001), while in the case of railroad tracks, agility relates primarily to the ability to overcome obstacles. Due to its physical features, the common toad was more likely to become stranded at the rail, indicating that this species is more vulnerable to railway mortality. However, other species that do not cross the track because of their small body size may also be affected by the railroad, but at the level of gene flow (Reh, 1989; Vos et al., 2001) which represents a conservation issue that is worthy of further study.

Further investigations examining in detail the effect of individual physical features on amphibian railroad mortality, railway-related migration behavior of amphibians, as well as gene flow among amphibian populations isolated by railway line, are warranted.

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